# This Page Is Inserted by IFW Operations and is not a part of the Official Record

## **BEST AVAILABLE IMAGES**

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images may include (but are not limited to):

- BLACK BORDERS
- TEXT CUT OFF AT TOP, BOTTOM OR SIDES
- FADED TEXT
- ILLEGIBLE TEXT
- SKEWED/SLANTED IMAGES
- COLORED PHOTOS
- BLACK OR VERY BLACK AND WHITE DARK PHOTOS
- GRAY SCALE DOCUMENTS

## IMAGES ARE BEST AVAILABLE COPY.

As rescanning documents will not correct images, please do not report the images to the Image Problems Mailbox.

Japanese Patent Office (JP)
Patent Application Laid Open
JAPANESE PATENT KOKAI (A) H6-122696

Published 6th May 1994

International Recognition Office Handling Classification Code Number

C 07 H 19/04

21/02

21/04

2

C 12 Q 1/68

A 7823-4B

Fl

Technical Display Location

Request for Examination: Not yet requested

Number of Claims: Six

Number of Pages in the Japanese Text: Thirteen

Application Number: H4-272914

Date of Application: 12th October 1992

Applicant: 000001007

Canon K.K.

30-2 Shimo Maruko-3-chome, Ota-ku, Tokyo-to,

Japan

Inventor: Y. Tomita

c/o Canon K.K., 30-2 Shimo Maruko-3-chome,

Ota-ku, Tokyo-to, Japan

Inventor: T. Mihara

c/o Canon K.K., 30-2 Shimo Maruko-3-chome,

Ota-ku, Tokyo-to, Japan

Inventor: H. Okamoto

c/o Canon K.K., 30-2 Shimo Maruko-3-chome,

Cta-ku, Tokyo-to, Japan

Agent: Patent Attorney T. Marujima

Title of the Invention:

Mononucleotide resembling dyes for DNA (RNA) probe
purposes, and DNA (RNA) probes

Abstract

Constitution:

Mononucleotide resembling dyes for DNA (RNA) probe purposes, characterized in that they contain dye, phosphoric acid radical and sugar, the sugar is ester bonded to from 1 to 3 phosphoric acid radicals or phosphoric acid radical derivatives and the said sugar is glycoside bonded to the said dye, DNA (RNA) probes to which the said dye is bonded, and a method of detecting DNA in which the said DNA (RNA) probes are used.

The dyes for DNA probe purposes of this invention can be synthesized in large amounts and they are water-soluble, and they can be introduced into a DNA probe easily and quantitatively. Furthermore, the DNA probes into which the said dyes have been introduced can be measured very accurately and they are of general utility which is not dependent on the base sequence of the nucleic acid which is to be detected.

Scope of the Patent Claims [Claim 1]

Mononucleotide resembling dyes for DNA (RNA) probe purposes, characterized in that they contain at least one dye selected from among the azo dyes, anthraquinone dyes, indigoid dyes, phthalocyanine dyes, carbonium dyes, quinoneimine dyes, methine dyes, quinoline dyes, nitro dyes, nitroso dyes, benzoquinone dyes, naphthoquinone dyes, naphthalimide dyes, penoline dyes and azulene dyes, phosphoric acid radical and sugar, the sugar is ester bonded to from 1 to 3 phosphoric acid radicals or phosphoric acid radical derivatives, and the said sugar is glycoside bonded with the said dye.

[Claim 2] Nucleotide resembling dye for DNA (RNA) probe purposes, according to Claim 1, characterized in that protective groups are added to the free hydroxy groups of the said sugar.

[Claim 3]

Nucleotide resembling dye for DNA (RNA) probe purposes, according to Claim 1 or Claim 2, with which the molar extinction coefficient ( $\mathcal{E}$ ) of the said dye is from  $1 \times 10^4$  to  $1 \times 10^6$ .

[Claim 4]

characterized in that probe, (RNA) DNA dye for DNA (RNA) probe mononucleotide resembling purposes as described in Claims 1 to 3 is bonded via a phosphoric acid radical to the 5' end and/or the 3' end a poly or oligonucleotide to form a poly or oligonucleotide.

[Claim 5]

DNA (RNA) probe, characterized in that a plurality of mononucleotide resembling dye units for DNA (RNA) probe purposes as described in Claims 1 to 3 are bonded via a phosphoric acid radical to the 5' end and/or the 3' end of a poly or oligonucleotide to form a poly or cligonucleotide.

[Claim 6]

Method for the detection of DNA, characterized in that, in a method for the detection of DNA in which a DNA probe and sample DNA are mixed and the DNA is detected by detecting a hybrid formed by the target DNA which is included in the sample DNA and the DNA probe, the said DNA probe is a DNA probe as described in Claim 4 or Claim 5.

Detailed Description of the Invention

[0001]

#### Industrial Field of Application

This invention concerns DNA (or RNA) probes for detecting the presence or absence of a specific base sequence in DNA or RNA, and the dyes used in the said probes.

[0002]

#### Prior Art

A DNA probe is a material in which some substance is attached as a label to DNA or a nucleotide which has a base sequence (one chain) which is complementary to the base sequence of the DNA which is being sought (the target DNA) for seeking a specified base sequence within DNA (an RNA probe is the same and so the case of DNA will be described hereinafter).

[0003]

In the past there were methods in which radioactive isotopes were used as labelling substances for DNA probes. With these, phosphorus, for example, was replaced with a radioactive isotope, and this was then detected using autoradiography or a Geiger counter for example.

[0004]

However, the methods in which radioactive isotopes are used are dangerous and require handling facilities for radioactive isotopes and equipment such as Geiger counters, and there is a further disadvantage in that they cannot be used for examinations within a human body.

### Pigure 1

## Figure 2

## Figure 5